PRODUCTION OF BIOETHANOL FROM KUNDUR [BENINCASA HISPIDA (THUNB.) COGN.]

SITI SALBIAH BINTI MUTALIB

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ABSTRACT

Bioethanol is a clean burning, renewable resources which has become one of the alternative biofuels which are environmentally friend. Bioethanol has been proved that it can be produced by using agro-industrial and food wastes. Kundur (Benincasa hispida) also known as fuzzy melon is a vegetable crop that popular in Asian for their nutritional and medical attributes such as anti-obesity, anti-inflammatory, anti-diarrheal, antioxidant and etc. The aim of the study is to investigate the production of bioethanol from Kundur. This study was conducted by using different parts of Kundur. The moisture contents in Kundur seeds, pulps and fleshes are 77, 86 and 97 % (w/w) respectively. Kundur fleshes have the highest amount of reducing sugar (glucose) and carbohydrate contents which is 2.909 and 79.885 g/L. Meanwhile the amount of reducing sugar and carbohydrate are 0.924 and 33.333 g/L for Kundur seeds and 0.796 and 13.103 g/L for Kundur pulps.

Kundur juices were selected for the batch fermentation in the shake flasks to investigate the suitability of Kundur juice to produce ethanol by using *Saccharomyces Cerevisae* as yeast. The fermentation process was carried out in duplicate at 30°C, pH 5.5 and 200rpm for 72hours. According to the results obtained, the ethanol yield for the fermentation process without nutrient supplement is in the range of -0.192~ -0.194 (g ethanol/g biomass) while the specific growth rate is in the range of -0.0148~ -0.015h^{-1}. As for the fermentation process with nutrient supplement, the ethanol yield and specific growth rate is in the range of -0.136~ -0.1442 (g ethanol/g biomass) and -0.017h^{-1}. Hence, we can conclude that Kundur juice is not suitable for ethanol production

PENGHASILAN ETHANOL DARIPADA KUNDUR [BENINCASA HISPIDA (THUNB.) COGN.]

ABSTRAK

Bioethanol adalah pembakaran bersih, salah satu sumber alternatif biofuel yang boleh diperbaharui dan mesra alam. Bioethanol telah dibuktikan bahawa ia boleh dihasilkan dengan menggunakan bahan buangan agro-industri dan makanan. Kundur (Benincasa hispida) juga dikenali sebagai tembikai kabur adalah tanaman sayur-sayuran yang popular di Asia dari sudut pemakanan dan perubatan seperti anti-obesiti, anti-radang, anti-cirit-birit antioksidan, dan lain-lain Tujuan kajian ini adalah untuk menkaji penghasilan bioetanol dari Kundur. Kajian ini telah dijalankan dengan menggunakan bahagian-bahagian Kundur. Kandungan lembapan dalam benih, kulit dan isi Kundur adalah 77, 86 dan 97% (w / w). Isi Kundur mempunyai jumlah tertinggi pegurangan gula (glukosa) dan karbohidrat iaitu 2.909 dan 79.885 g/L. Sementara itu, jumlah pengurangan gula (glukosa) dan karbohidrat dalam biji Kundur adalah 0.924 and 33.333 mg/L manakala 0.796 dan 13.103 g /L untuk kulit Kundur.

Jus Kundur telah dipilih dalam fermentasi untuk menyiasat kesesuaian jus Kundur bagi menghasilkan etanol dengan menggunakan Saccharomyces Cerevisae sebagai yis. Proses fermentasi telah dijalankan dalam dua salinan pada 30 ° C, pH 5.5 dan 200rpm selama 72jam. Menurut keputusan yang diperolehi, hasil etanol untuk proses fermenatsi tanpa tambahan nutrien adalah dalam julat -0,192 ~ -0,194 (g etanol / g biojisim) manakala kadar pertumbuhan spesifik adalah dalam julat -0,0148 ~-0.015h-1. Manakala untuk proses fermentasi dengan tambahan nutrien, hasil etanol dan kadar pertumbuhan spesifik adalah dalam julat -0,136 ~ -0,1442 (g etanol / g biojisim) dan-0.017h-1. Oleh itu, kita boleh menyimpulkan bahawa jus Kundur tidak sesuai untuk pengeluaran etanol.

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LIST OF SYMBOLS

CO_2	-Carbon Dioxide
CO	-Carbon Monoxide
HCl	-Hydrochloric Acid
RPM	- Rotation per Minutes
KJ	-Kilo Joules
%	- Percent
%w/w	- Percent Gram over Gram
g	- Gram
Tg	-Tera Gram
mg	-Milli Gram
L	- Liter
GL	-Giga liter
mL	-Milli liter
OD	- Optical Density
nm	-nano Meter

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CHAPTER 1

INTRODUCTION

1.1 Research Background

Bioethanol is an ethanol (ethyl alcohol) produced from plants such as soybeans, sugarcane, grass and wood. Several researchers have successfully produced ethanol employing juice or pulp from various fruits such as guava (Bardiya et al., 1974; Dhawan et al., 1983), pineapple, blackberry, orange, tamarind, cashew fruit (Maldonado *et al.*, 1975), banana (Kundu et al., 1976; Srivastava, 1984) and chashew apple (Modi, 1984; Sandhu and Joshi, 1994). Nowadays, bioethanol along with biodiesel has become one of

the most promising biofuels and is considered as the alternative medium to fossil transport fuels in Europe and in the wider world.

Bioethanol is seen as a good alternative fuel due to the source crops can be renewably and inexpensive. The economics of ethanol production are significantly influenced by the cost of the raw materials, which account for more than half of the production costs (Classen et al., 1999). This is important to achieve a lower production cost. Therefore, in order to achieve a lower production cost, the raw materials supply must be cheap. Moreover, the use of bioethanol is generally CO_2 neutral. This is because in the growing phase of the source crop, CO_2 is absorbed by the plant and oxygen is released in the same volume that CO_2 is produced in the combustion of the fuel (Wyman, 1990; Chandel et al., 2006). In addition, the emission and toxicity of ethanol are lower than those of fossil fuels such as petroleum (Wayman and Hinman, 1990).

1.2 Problem Statement

In recent years, most country around the world seeking a solution for the uncertain fuel supply and efforts to reduce carbon dioxide, CO_2 emission. This is because the availability of fossil fuels sources nowadays can only survive for another 20 to 30 years. Besides that, the use of fossil fuels such as coal, gasoline and natural gas has increased the concentration of carbon dioxide in the environment due to combustion

process. The increase of carbon dioxide concentration in the environment will lead to the increase in the earth's temperature which then will cause global warming. So that, to encounter this problems the solution should be developed.

Recently several resources had succeed produce ethanol from various fruits and plants such as rice straw, corn stalk (Kuhad and Singh, 1993), pine, aspen (Olsson and Hagerdal, 1996) and etc as an alternative fuel. Therefore, the present study was undertaken to produce ethanol by using Kundur fruit.

1.3 Research Objectives

The main objectives are:

(i) To investigate the properties of Kundur.

(ii) To identify the suitability of Kundur in ethanol production.

(iii) To identify the kinetic parameter of fermentation process.

1.4 Research Scopes

According to the objectives stated, the scope of research is to investigate the properties of Kundur parts consists of seed, flesh and pulp. The properties that will be

investigates are moisture contents, carbohydrates and reducing sugars. Apart from that, the purpose of this study is to identify the suitability of Kundur as a raw material for producing ethanol. In addition, this study will also investigate the kinetic parameter of the fermentation process which is biomass generation rate, glucose consumption rate and ethanol production rate

CHAPTER 2

LITERATURE REVIEW

2.1 Ethanol

Ethanol having a formula C_2H_5OH (or empirically C_2H_6O) is a second member of the aliphatic alcohol series which is a group of chemical compounds, whose molecules contain a hydroxyl group, –OH, bonded to a carbon atom. Ethanol also known as ethyl alcohol, ethyl hydroxide and etc is a clear, colorless, and volatile liquid with a characteristic of agreeable odor (Pradyot Patnaink, 2007). The main physical properties of ethanol are as shown in **Table 2.1**. In a dilute aqueous solution, ethanol has

a somewhat sweet flavor, but in more concentrated solutions it has a burning taste. Other physical property of ethanol is that it is miscible (mixable) in all proportions with water and with most organic solvents, with quite an affinity for moisture absorption, even from the air.

Table 2.1 Physical Properties of Ethanol

Component	
Formula weight	46.06g/mol
Boiling point	78.5°C
Melting point	-114.1°C
Density	0.789g/ml at 20°C
Vapor pressure	43 torr at 20°C
Latent heat of evaporation	396 BTU/lbm
Gravimetric lower heating value	11,604 BTU/lbm

Source: CRC Handbook of Chemistry and Physics, (1993)

2.2 Applications of Ethanol

The largest use of ethanol is as a motor fuel in the automotive fuel industry. Ethanol is used as an alternative vehicle fuel, for example as E85 which is a mixture of

85% ethanol and 15% of gasoline by volume. For E85 fuel, 100 km driven consumes 2.2L of gasoline and 12L of bioethanol (S. Kim and B.E. Dale, 2004). Therefore 1L of bioethanol could replace 0.72L of gasoline. The market for this sector has highest growth potential. Many countries have attempted to replace some of their gasoline consumption with ethanol by mixing certain percentage of ethanol into gasoline to achieve the resulting product called gasohol. The largest national fuel ethanol industries exist in Brazil. Ethanol for fuel use is further divided into hydrous and anhydrous alcohol, each contained different purity from distillation.

Besides that, ethanol is useful as a solvent for many substances and in making perfumes, paints, lacquer, and explosives (Pradyot Patnaink, 2007). Alcoholic solutions of nonvolatile substances are called tinctures while if the solute is volatile, the solution is called a spirit. Ethanol acts as a drug affecting the central nervous system. Its behavioral effects stem from its effects on the brain and not on the muscles or senses themselves. It is a depressant, and depending on dose which can be a mild tranquilizer or a general anesthetic. It suppresses certain brain functions. At very low doses, it can appear to be a stimulant by suppressing certain inhibitory brain functions. However, as concentration increases, further suppression of brain functions produce the classic symptoms of intoxication which are slurred speech, unsteady walk, disturbed sensory perceptions, and inability to react quickly. Meanwhile, at very high concentrations, ethanol produces general anesthesia which is a highly intoxicated person will be asleep and very difficult to wake, and if awakened, he unable to move voluntarily.

2.3 Alternative Sources of Fuels

Energy demand increase year by years. According to BP statistical review of world energy June 2011, the consumption of energy growth reached 5.6%, the highest rate since 1973 in 2010. The total consumption of energy in 2010 easily surpassed the pre-recession peak reached in 2008. Consumption growth accelerated in 2010 for all regions. China energy consumption grew by 11.2%, and china surpassed the US as the world's larger energy consumption. Globally, energy consumption grew more rapidly than the economy, which means the energy intensity of economic activity increase for a second consecutive year. Therefore, we need an alternatives fuel to sustain the energy demand. Ethanol has already been introduced on a large scale in Brazil, USA, and some European countries, and it is expected to be one of the dominating renewable biofuels in the transport sector within the coming 20 years.

Ethanol can be blended with petrol or used as neat alcohol in dedicated engines that taking advantage of higher oxygen content, higher octane number, higher heat of vaporization and reduction of CO emission (Cardona et al., 2010). Furthermore, it is an excellent fuel for future advance flexfuel hybrid vehicles. Nearly, all fuel ethanol is produced by fermentation of sucrose in Brazil or corn glucose in the USA. However these raw material bases will not be sufficient to satisfy the international demand. Therefore many countries had made many researches to produce bioethanol from other sources such as oat, rice, sorghum and etc. The raw materials for production of bioethanol can be from agriculture residues, virgin biomass, waste paper, organic

fraction of municipal solid waste (MSW) and other materials containing fermentable sugar (McMillian, 1997).

Table 2.2 Quantities of Wasted Crop Potentially Available for Bioethanol Production

	Africa	Asia	Europe	North America	Central America	Oceania	South America	Subtotal
Corn (Tg)	3.12	9.82	1.57	0.30	1.74	0.01	4.13	20.70
Barley (Tg)	0.17	1.23	2.01	0.01	0.01	0.19	0.04	3.66
Oat (Tg)	0.004	0.06	0.43	0.01	0.001	0.001	0.05	0.55
Rice (Tg)	1.08	21.86	0.02	0.96	0.08	0.02	1.41	25.44
Wheat (Tg)	0.83	10.28	4.09	0.02	0.24	0.82	0.91	17.20
Sorghum (Tg)	2.27	0.54	0.004	0.00	0.13	0.001	0.18	3.12
Sugar cane (Tg)	0.46	1.64	0.00	0.00	0.36	0.00	0.74	3.20
Subtotal (Tg)	7.94	45.43	1.30	1.30	2.56	1.05	7.45	73.86

Source: S. Kim, B.E. Dale, (2003)

Table 2.3 Potential of Bioethanol Production from Waste crop

	Africa	Asia	Europe	North America	Central America	Oceania	South America	Subtotal
Corn (GL)	2.17	6.82	1.09	0.21	1.21	0.01	2.87	14.4

Table 2.3 Continue

Barley (GL)	0.12	0.83	1.35	0.005	0.01	0.13	0.03	2.46
Oat (GL)	0.002	0.04	0.30	0.01	0.0004	0.001	0.03	0.38
Rice (GL)	0.71	14.4	0.02	0.63	0.05	0.02	0.93	16.8
Wheat (GL)	0.55	6.78	2.70	0.02	0.16	0.54	0.60	11.3
Sorghum (GL)	1.55	0.37	0.003	-	0.09	0.0004	0.12	2.14
Sugar cane (GL)	0.23	0.82	-	-	0.18	0.0001	0.37	1.59
Subtotal (GL)	5.33	30.1	5.45	0.87	1.70	0.70	4.95	49.1

Source: S. Kim, B.E. Dale, (2003)

2.4 Kundur [Benincasa hispida (Thunb.) Cogn.]

Kundur which is also known as fuzzy or hairy melon is also called Chinese squash or moqua. Mature fruits of some *Benincasa hispida* [syn. *B. cerifera* (Thunb) Cogn.] are commonly called wax gourd, winter gourd or Chinese preserving melon. *Benincasa hispida* can be stored for a long period of time which is for many months. The immature Kundur fruits have a delicious flavor, stronger and distinctive. Flavor can change during storage, with the fuzzy melon taking on an acidic, less agreeable flavor.

Kundur fruit is an important source of water–soluble polysaccharide. Polysaccharides that present in Kundur juice are mainly arabinogalactans (Mazumder, Ray, and Ghosal et al, 2001). Natural sugars that are present in immature and mature Kundur fruit pulp are glucose and fructose. The level of both glucose and fructose are reported to be reduced from 0.9% to 0.5% and 0.8% to 0.5% respectively, as the fruit matured (Wills, Wong, Scriven and Greenfield et al, 1984).

Table 2.4 Nutritive Composition of Kundur per 100 g Edible Portion

Component	*Benincasa hispida*
Edible portion (%)	98
Water (%)	93.8
Energy (KJ)	44
Protein (g)	0.7
Fat (g)	0.1
Carbohydrate (g)	2.0
Dietary fiber (g)	2.1
Organic acids (g)	0.04
Ash (g)	0.7
Minerals	
Ca (mg)	12
K (mg	250
Mg (mg)	15

Fe (mg)	0.3
Na (mg)	2

Table 2.4 Continue

Zn (mg)	0.2
Vitamins	
A (mg)	0.02
thiamin (mg)	0.07
riboflavin (mg)	0.05
Niacin (mg)	0.20
C (mg)	69

Source: Marita Cantwell, Xunli Nie, Ru Jing Zong, and Mas Yamaguchi, (1996)

The diversity and antiquity of cultivars in China suggest that this crop may be indigenous to southern China (Yang and Walters, 1992). Cultivars of *B. hispida* are classified on the basis of wax formation on the mature fruit, shape, fruit size and pubescence of the immature fruits which is from which its common name is derived. Generally, separate cultivars are used for immature vegetable production and for mature fruit production (Yang and Walters, 1992). One vegetable type has fruits which are cylindrical and roundish with many bristle-like trichomes on the epidermis. The other jointed gourd has immature fruits which are narrowed in the center that have dumbbell shaped with their length 2-3 times their width.